



Related Functions of Signaling Proteins in Maize Stomata & Arabidopsis Roots

Erika Norris, Michelle Facette

Department of Biology, UMass Amherst

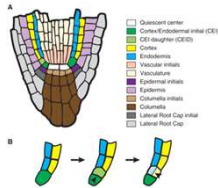


United States Department of Agriculture
National Institute of Food and Agriculture

Background & Objectives

ZmPAN1, AtPNL1, & AtPNL2 are signaling molecules in maize & Arabidopsis

- Signaling molecules for cell division
- Currently it is unknown how they signal



PAN1 is a protein found in maize

- Required for making stomata & required for asymmetric cell division

pnl1* & *pnl2* are found in *Arabidopsis thaliana

- Are **not** needed for stomata in Arabidopsis, but are required in the root & help to form the cell plate during division
- However, the *pnl* mutants also show abnormal cell division within the roots

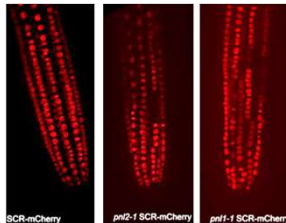
Figure 1: Root tips of Arabidopsis, circles highlight abnormal division sites



How do PAN1, PNL1, & PNL2 relate?

- Orthologs; come from the same distant relative
- Studying the *pnl* mutants can lead to an understanding of the PAN mutant in maize

Figure 2: *pnl* mutations within the roots of Arabidopsis.



These cell types are missing in the mutants, are there any other cells that are missing?

In this study, I looked at the *pnl* mutants in Arabidopsis, focusing on the root hairs & the columella stem cells in the root tip

Methods

- Col-0, *pnl1*, *pnl2* plated on 1/2 MS Media containing 1% agar & 1% sucrose
- Root hairs imaged using a light microscope
- Plants were placed into FAA for a few hours, then washed twice with 1X PBS buffer before Clearsee (CS) was added
 - In CS for at least 1 day, up to 1 week
- Plants were washed in decreasing concentrations of CS until able to withstand 100% H₂O
- Stained plants in Lugol's iodine solution for 2 min
- Washed plants briefly in H₂O, then washed again for 8 min
- Plants were wet mounted using H₂O & observed using a Nikon Eclipse TE2000-S microscope

Acknowledgements

I would like to thank the CAFE Summer Scholars program for the opportunity to conduct this research. I would also like to thank Michelle Facette & the Facette Lab members, Le Liu, Nia Gray, & Savannah Metzger, for their ongoing guidance & counsel, as well as Arif Ashraf for continuous support.

Results

Are *pnl1* & *pnl2* required for only some division (endodermal) or all division?

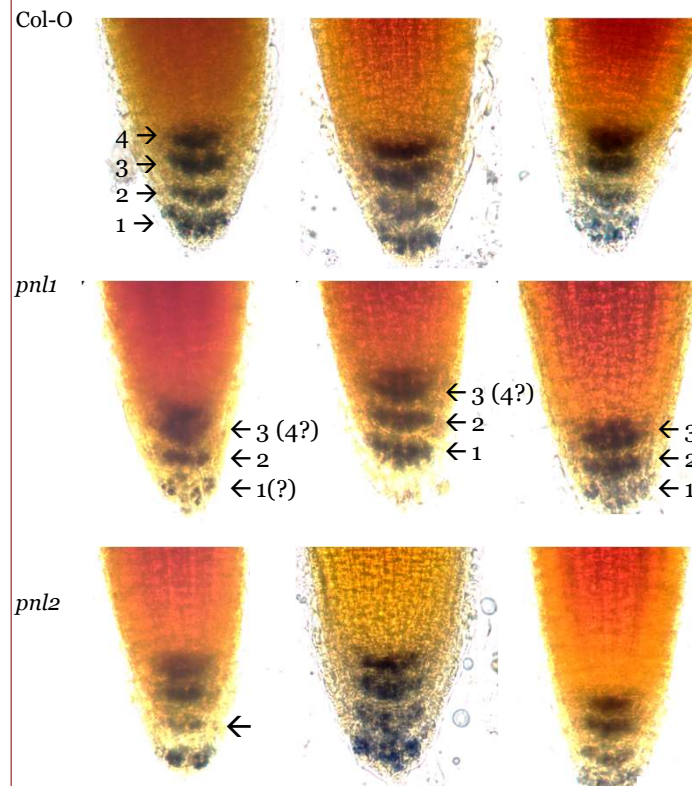


Figure 3: Stained starch within the columella cells in the root tips of Arabidopsis.

Preliminary imaging suggests that *pnl1* and *pnl2* have root cap specification defects.

Double blind quantitative analyses are pending after further data collection.

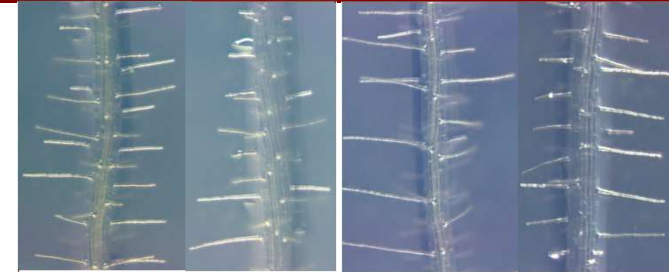


Figure 4: Col-0 root hairs in the middle of root & at the tip

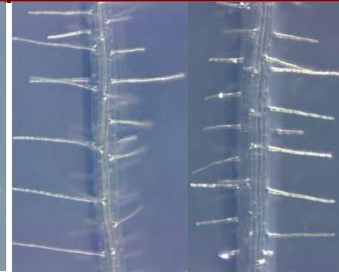


Figure 5: *pnl1* root hairs in the middle of root & at the tip

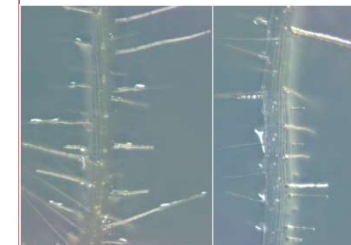


Figure 6: *pnl2* root hairs in the middle of root & at the tip

The images show that there is not an observable difference of root hair spacing amongst the genotypes studied.

Impact & Applications

-What are PAN1, *pnl1*, & *pnl2* doing?

-Image quality issues because of using Lugol's staining versus chloral hydrate?

-No observable difference in root hairs between control & mutants. Potential further testing.

References

- Hong JH, et al (2015) *Front. Plant Sci.* 6:206. doi: 10.3389/fpls.2015.00206
- Clark, L. (2010). PNL1 & PNL2 : Arabidopsis homologs of maize PAN1. *UC San Diego*. ProQuest ID: Clark_ucsd_0033M_11094. Merritt ID: ark:/20775/bb43698743.